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The relationship of RS645439735, RS66142748 and RS670830003 mutations in the PIT-1 gene to the productive and reproductive performance of local goat

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Abstract---The current study included 85 does with the same management and feeding conditions belonging to two breeds (Local and Shami goats), located in the west of Baghdad. 4–5 years old, multiparous, lactating and in their first to fourth lactation. PIT-1 gene polymorphism was indicated and results showed a significant effect ($P \leq 0.05$) PIT-1 gene polymorphism (rs645439735) on protein rate in Cyprus breed, the highest value was recorded in the individuals with hetero genotype (AC) namely 3.16 % compared with 2.92 and 2.80% in both wild and mutant genotypes respectively. Results showed a significant effect ($P \leq 0.05$) of rs661742748 SNP on some of milk traits in Cyprus breed, the milk peak was higher in hetero genotypes compared with the wild type namely, 64.0 and 34.46 day respectively. Fat rate was increased significantly in hetero type (5.6%) compared with wild type (3.11%). Results showed a significant effect of rs670830003 SNP on weaning weight and body Weight gain from birth to weaning in the local goat breed, the highest values were 16.83 and 14.04 Kg was recorded in individuals with hetero genotype (TG) while the lowest values were in individuals with the wild type namely, 13.81 and 11.18 Kg respectively. No significant effect of this SNP on the milk traits and fertility in Cyprus goat breed.

Keywords--- PIT-1 gene, Local and Cyprus goat, production, fertility.

Introduction

Goat is one of ancient animal species which domesticated before 10500 years ago in Mesopotamia and then widespread to another area around the world (AL-Samari et.al.,2016; Amillset.al., 2017; Alberto et.al, 2018; Gipson, 2019). Goat is one of the most animal species that good acclimatized towards harsh

environmental conditions and poor pastures compared with cattle or sheep (Ibtisham et. al., 2017). Goats are of high genetic value, breeding does not need high cost and its management is simple and more resistant to epidemic diseases and parasites of sheep and cows, which made breeding more suitable for drought and low-yield agricultural areas (AL-Jorani et.al., 2020). The classic selection of domestic animals such as goat lead to increase the performance of individuals and the current studies consists on marker assisted selection as indirect selection and determine the quantitative trait loci (QTL) for economic traits such as reproductive and productive important traits (Coli, 2018).

The increase demands on animal's products and wide gap between production and consumption make man thinking of new methods to improve the animal performance and one of these methods is candidate genes such as pituitary transcription factor (PIT-1) which located on first chromosome and contains of six exons with total volume 21000 bp (Daga et. al., 2013). Many studies reported that the PIT-1 gene is related strongly with the gene expression of growth hormone and another important hormones such prolactin and TSH β (Alakilli, et.al., 2012; Shweta et.al., 2013 ; Sobreier et. al., 2016). The major aim of the current study is to determine the polymorphism of PIT-1 gene and exploit the genetic variance resulted from the single nucleotide polymorphism (SNPs) as a tool of indirect selection to improve the local goat performance in Iraq under the farming conditions.

Materials and Methods

The current study included 85 does with the same management and feeding conditions belonging to two breeds (Local and Shami goats), located in the west of Baghdad. 4–5 years old, multiparous, lactating and in their first to fourth lactation.

Sampling

A jugular blood sample was collected from each goat using vacuum tubes with EDTA k2 as an anti coagulant. Protein Kinase was used to isolate DNA. PIT-1 gene polymorphism was determined using forward 5-CATCCCTATATCGTCCCTAGTAT-3' and reverse 5- GAGCGCATGCTGGTATTT -3' primers (FAO, 2011). Cyclic applied were denaturation 94°C/5 minutes followed by 30 cycles 94°C/30 seconds, primer annealing 58°C/30 seconds PCR product synthesis. The PCR products were separated electrophoretically in 1.5% agarose gel. Special kit was used manufactured by Bioneer company (Republic of South Korea). Growth trait such as birth weight, weaning weight, live animal weight and total gain from birth to weaning were recorded. Body dimensions such as body length, high and heart girth were measured by using a tape included the following traits:

- Body weight at birth and weaning for all individuals was weighted by high accuracy balance.

- Heart Girth (HG) is circumferential measure taken around the chest, Withers Height (WH): This is the distance from the surface of a platform to the withers,
- Rump Height(RH):This is the distance from the surface of a platform to the rump,
- Body Length(BL):This is the distance from occipital protuberance to the base of the tail. (Simeon and Adewale, 2017).

Statistical analysis

Data were analyzed using SAS (2012) computer program by general linear model procedure (GLM) according to the following model:

$$Y_{ijk} = \mu + T_i + e_{ijk}$$

Where: μ : the overall mean

T_i : effect of PIT-1 genotypes

e_{ijk} : is a random error.

Significant differences among groups were detected by general linear model (GLM) and Duncan's multiple range test was used to compare differences among means.

Results and Discussion

Table 1
Effect of rs645439735 SNP on local and Cyprus performance

Breed	traits	AA	AC		p-value
		28	6		
Local	Birth w (kg)	2.6± 0.14	2.63±0.40		n.s
	W W (kg)	0.67±14.58	14.5±1.28		n.s
	Gain (kg)	11.91± 0.6	11.8±0.97		n.s
	Bw live (cm)	43.14± 1.56	37.66±2.26		n.s
	H g (cm)	83.71±1.02	80.0±2.25		n.s
	Bl (cm)	83.92± 0.95	79.33±2.2		n.s
	Hf (cm)	77.14± 0.53	75.66±1.25		n.s
	Hr (cm)	79.39±0.53	77.16±1.83		n.s
	Fertility (%)	0.66±0.08	0.66±0.21		n.s
Cyprus		AA	AC	CC	
		46	7	2	
	Birth w (kg)	2.36±0.11	2.5±0.16	2.15±0.35	n.s
	W W (kg)	15.34±0.51	14.42±1.06	12.0±0	n.s
	Gain (kg)	12.98±0.47	11.92±0.94	9.85±0.35	n.s
	Bw live (cm)	46.45±1.72	50.85±3.96	41.5±6.5	n.s
	H g (cm)	84.63±1.14	87.28±2.63	81.5±3.5	n.s
	Bl (cm)	82.86±0.96	84.14±2.36	76.5±3.5	n.s
	Hf (cm)	76.54±0.68	78.28±1.98	78.5±4.5	n.s

	Hr (cm)	78.76±0.68	80.57±2.09	80.5±4.5	n.s
	Fertility (%)	0.78±0.06	0.85±0.14	1.00±0	n.s

Results showed no significant effect of rs645439735 SNP on all traits that studied and the results indicated that a two genotypes in local goat breed (AA and AC) while three genotypes were recorded in Cyprus breed namely AA (wild), AC (Hetero) and AC (Mutant).

Table 2
Effect of rs645439735 SNP on local and Cyprus performance

Breed	traits	AA	AC		p-value
		20	4		
Local	Twining (kid)	1.7± 0.14	2.0±0		n.s
	TMY (kg)	239.53±27.77	204.57±69.05		n.s
	Lac – p (day)	226.6±8.62	189.25±39.30		n.s
	Peak(day)	37.7± 1.93	46.75±4.49		n.s
	Fat (%)	2.96±0.27	2.91±1.10		n.s
	Pro (%)	3.0±0.04	3.07±0.11		n.s
	Lactose (%)	4.47±0.05	4.52±0.21		n.s
	SNF (%)	8.2±0.08	8.45±0.38		n.s
Cyprus		AA	AC	CC	
		36	6	2	
	Twining (kid)	1.75±0.08	1.66±0.33	2.0±0	n.s
	TMY (kg)	219.77±18.77	252.35±53.88	155.6±28.6	n.s
	Lac – p (day)	193.58±7.78	199.16±16.21	203.5±12.5	n.s
	Peak(day)	34.66±1.53	38.66±2.92	33.0±2.0	n.s
	Fat (%)	3.13±0.19	3.6± 0.28	2.47±0.47	n.s
	Pro (%)	2.92±0.03aab	3.16±0.09a	2.8±0.25b	*
	Lactose (%)	4.34±0.04	4.51±0.14	4.3±0.37	n.s
	SNF (%)	8.01±0.06	8.29±0.23	7.91±0.67	n.s

Results showed a significant effect ($P \leq 0.05$) PIT-1 gene polymorphism (rs645439735) on protein rate in Cyprus breed, the highest value was recorded in the individuals with hetero genotype (AC) namely 3.16 % compared with 2.92 and 2.80% in both wild and mutant genotypes respectively (Table-2). No significant effect was noticed in other traits according this SNP polymorphism. Results showed a significant effect ($P \leq 0.05$) of rs661742748SNP on body dimensions in local breed (Table-3). The highest values of High , , Hf and Hr were recorded in mutant genotype namely, 95, 83 and 85 cm respectively compared with 83.1 , 76.86 and 78.96 cm in the individuals of local breed and wild type (CC) respectively. In Cyprus breed, there were no significant effect of this SNP on body dimensions and no any effect was recorded on fertility in both local or Cyprus breeds according this SNP.

Table 3
Effect of rs661742748 SNP on local and Cyprus performance

Breed	traits	CC	CT	TT	p-value
		31	4	1	
Local	Birth w (kg)	2.6±0.14	2.87±0.42	3.6 ±0	n.s
	W W (kg)	13.93±0.54	18.87±2.56	17.0 ±0	n.s
	Gain (kg)	11.32± 0.43	16.0±2.68	±013.4	n.s
	Bw live (cm)	41.86±1.42	41.0±5.11	±056.0	n.s
	H g (cm)	83.1±0.98b	79.75±2.05b	95.0±0a	*
	Bl (cm)	82.86± 0.95	83.25±3.54	90.0±0	n.s
	Hf (cm)	76.86±0.52b	75.5±0.95b	83.0±0a	*
	Hr (cm)	78.96±0.59ab	77.75±0.94b	85.0±0a	*
	Fertility (%)	0.61± 0.08	1.00±0	1.00±0	n.s
Cyprus		CC	CT		
		53	2		
	Birth w (kg)	2.37±0.09	2.3±0.3		n.s
	W W (kg)	15.11±0.47	15.0±0		n.s
	Gain (kg)	12.73±0.43	12.7±0.3		n.s
	Bw live (cm)	47.24±1.55	36.0±9.0		n.s
	H g (cm)	85.15±1.02	77.0±5.0		n.s
	Bl (cm)	82.9±0.89	80.0±6.0		n.s
	Hf (cm)	76.92±0.65	74.5±1.50		n.s
	Hr (cm)	79.15±0.66	76.5±1.5		n.s
	Fertility (%)	0.81±0.05	0.50± 0.5		n.s

Table 4
Effect of rs661742748 SNP on local and Cyprus performance

Breed	traits	CC	CT	TT	p-value
		19	4	1	
Local	Twining (kid)	1.68± 0.15	2.0± 0	2.0 ±0	n.s
	TMY (kg)	225.71±30.65	240.62±36.73	358.05 ±0	n.s
	Lac - p (day)	219.15±12.09	223.75±10.07	230.0±0	n.s
	Peak(day)	40.26±2.10	36.75±4.83	29.0±0	n.s
	Fat (%)	3.01±0.34	2.4±0.21	4.0±0	n.s
	Pro (%)	3.02± 0.04	2.92±0.06	3.2±0	n.s
	Lactose (%)	4.48±0.06	4.37±0.07	4.8±0	n.s
	SNF (%)	8.25±0.10	8.03± 0.16	8.8±0	n.s
Cyprus		CC	CT		
		43	2		
	Twining	1.76±0.08	±01.0		n.s

	(kid)				
	TMY (kg)	223.17±17.3	±0140.5		n.s
	Lac - p (day)	195.04±6.84	±0184.0		n.s
	Peak(day)	34.46±1.16b	64.0±0a		*
	Fat (%)	3.11±0.16b	±0a5.6		*
	Pro (%)	2.95±0.03	±03.1		n.s
	Lactose (%)	4.36±0.04	±04.6		n.s
	SNF (%)	8.04±0.07	±08.39		n.s

Results showed a significant effect ($P \leq 0.05$) of rs661742748 SNP on some of milk traits in Cyprus breed (Table-4) the milk peak was higher in hetero genotypes compared with the wild type namely, 64.0 and 34.46 day respectively. Fat rate was increased significantly in hetero type (5.6%) compared with wild type (3.11%). All milk components did not differ significantly in local breed according this SNP.

Table 5
Effect of rs670830003 SNP on local and Cyprus performance

Breed	traits	TT	TG		p-value
		26	9		
Local	Birth w (kg)	2.62±0.16	2.78±0.24		n.s
	W W (kg)	13.81±0.6b	16.83±1.32a		*
	Gain (kg)	11.18±0.48b	14.04±1.30a		*
	Bw live (cm)	42.24±1.63	42.0±2.78		n.s
	H g (cm)	83.52±1.10	81.77±1.93		n.s
	Bl (cm)	83.2±1.08	82.88±1.79		n.s
	Hf (cm)	77.08±0.57	76.33±1.04		n.s
	Hr (cm)	79.16±0.66	78.55±1.0		n.s
	Fertility (%)	0.59±0.09	0.88±0.11		n.s
Cyprus		TT	TG	GG	
		51	3	1	
	Birth w (kg)	2.3±0.10	2.53±0.29	3.0 ±0	n.s
	W W (kg)	15.07±0.49	14.66±0.33	±018.0	n.s
	Gain (kg)	12.72±0.44	12.13±0.59	±015.0	n.s
	Bw live (cm)	46.8±1.58	43.66±9.26	±058.0	n.s
	H g (cm)	84.76±1.02	83.33±6.96	±094.0	n.s
	Bl (cm)	82.76±0.91	83.66±5.04	±082.0	n.s
	Hf (cm)	76.78±0.66	77.66±3.28	±077.0	n.s
	Hr (cm)	79.0±0.6	79.66±3.28	80.0±0	n.s
	Fertility (%)	0.82±0.05	0.33±0.33	1.0±0	n.s

Results represented in table-5 showed a significant effect of rs670830003 SNP on weaning weight and body Weight gain from birth to weaning in the local goat breed, the highest values were 16.83 and 14.04 Kg was recorded in individuals with hetero genotype (TG) while the lowest values were in individuals with the

wild type namely, 13.81 and 11.18 Kg respectively. No significant effect of this SNP on the milk traits and fertility in Cyprus goat breed.

Table 6
Effect of rs670830003 SNP on local and Cyprus performance

Breed	traits	TT	TG		p-value
		16	8		
Local	Twining (kid)	1.68±0.17	1.87±0.12		n.s
	TMY (kg)	247.02±33.61	207.08±36.54		n.s
	Lac - p (day)	220.37±13.31	220.37±12.61		n.s
	Peak(day)	39.56±2.15	38.5±3.81		n.s
	Fat (%)	2.95±0.38	2.94±0.39		n.s
	Pro (%)	3.02±0.04	3.0±0.08		n.s
	Lactose (%)	4.5±0.06	4.45±0.08		n.s
	SNF (%)	8.26±0.12	8.2±0.14		n.s
Cyprus		TT	TG	GG	
		42	1	1	
	Twining (kid)	1.7±0.08	1.0 ±0	2.0 ±0	n.s
	TMY (kg)	221.41±17.62	140.5±0	±0297.0	n.s
	Lac - p (day)	194.16±6.95	±0184.0	±0232.0	n.s
	Peak(day)	34.5±1.19b	64.0±0a	33.0±0b	*
	Fat (%)	3.08±0.16	±05.6	±04.3	n.s
	Pro (%)	2.94±0.03	3.1±0	±03.1	n.s
	Lactose (%)	4.35±0.046	4.6±0	±04.8	n.s
	SNF (%)	8.03±0.07	8.39±0	±08.42	n.s

Results indicated no significant effect of rs670830003 SNP on all milk traits in the local breed and also in Cyprus breed except lactation period which differed significantly ($P \leq 0.05$) to increase to 64.0 day in the individuals with hetero genotype compared with 34.5 and 33.0 day in both wild and mutant respectively. Many studies referred to a crucial difference in economic traits resulted from the genotypes variation of PIT-1 gene in many breeds of goat therefore, we can consider this gen as a candidate and important gene when we intend to select the best individual to improve the local goat performance. Feng et.al.(2013) referred that the polymorphism of this gene correlated significantly with fertility in JiningGrey goat breed while Zhu et.al. (2019) reported that the polymorphism of this gene effected on fertility and body dimensions in Shaanbei White Cashmere.Raziye and Güldehen (2019) reported that the polymorphism in the 6thexon region effected on body weight and fertility in Sanin goat breed while Siddiki et.al. (2020) reported that the multi allele in this gene which located in 5th exon region leads to significant effect on growth traits. The current results were agree with many of past studies results which referred to no significant effect of PIT-1 gene polymorphism on milk traits or growth traits such as Sarma et.al (2019).

Conclusion

the polymorphism of PIT-1 gene can a considerable tool to indirect selection because it an indicator for the genetic variation of many economic traits and finally we can depend on this gene confidently to put a selection plan to improve the local goat breeds in many directions such as fertility, milk production and meat production.

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